BIOLOGICAL NOTES ON MEXICAN (COLEOPTERA: LUCANIDAE) *LUCANUS (PSEUDOLUCANUS) MAZAMA* (LECONTE)[1]

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ABSTRACT: *Lucanus (Pseudolucanus) mazama* (LeConte 1861) lives in Mexican *Quercus* (oak) forests in decaying logs, where both adults and larvae are commonly found. The species is distributed in mixed pine-oak forests of the Sierra Madre Occidental, in areas of the Mexican states of Chihuahua and Sonora. The male reproductive apparatus is composed by 2 testicles, each with 12 testicular follicles, and 2 vas deferentia, 2 accessory glands, an ejaculatory bulb, and an ejaculatory duct. In females, the reproductive apparatus consists of 2 ovaries, each with 12 ovarioles, and 2 lateral oviducts, the common oviduct, bursa copulatrix, spermatheca with its gland, and vagina.

KEY WORDS: *Lucanus (Pseudolucanus) mazama*, Coleoptera, Lucanidae, México.

The Lucanidae family is a relatively small group of Scarabaeoidea, distributed widely around the world. Six Mexican species have been recorded (Maes 1992), representing about 0.5 percent of species known worldwide. Most of these species are endemic and known only on the basis of one or two adult specimens, since they are often difficult to collect. *Lucanus (Pseudolucanus) mazama* (LeConte 1861) is a Mexican stag beetle species that is large and relatively abundant. Although it was described from the United States, where it is found in Arizona, Colorado, New Mexico, and Utah (Maes 1992), it was recorded by Parry (1875) in northern Mexico, in the states of Chihuahua (Bates 1889, Villada 1901) and Sonora (Benesh 1944).

Stag beetle reproductive systems have been studied in few species, most of them European. Bordas (1900) described the reproductive apparatus of males of *Dorcus parallelipipedus* (Linné 1735) and *Lucanus cervus* (Linné 1758); the latter was illustrated by Franciscolo (1997), as was the typical scheme of the female Dorcinae. The only North American species known in this regard is the male *L. capreolus* (Linné 1763), described by Williams (1945). In a comparative study of the number of ovarioles (per ovary) in Scarabaeoidea, Ritcher and Baker (1974) observe that this number varies among different subfamilies of Lucanidae, from 6-6 in Aesalinae and Platycerinae, to 12-12 in Sinodendroninae and Lucaninae. Holloway (1960, 1998) studied numerous morphological structures of Lucanidae adults, and, considering the complete cuticular structures of the male and female genitalia, concluded that they "exhibit characters of high taxonomic and phylogenetic value."

Precise information had previously been lacking on the distribution of Mexican *L. mazama,* its habits, larval development, and male and female reproductive anatomy. These subjects are dealt with in the present paper.

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METHODS

This study examined 20 Mexican *L. mazama* adults from the collections of the Instituto de Ecología, A.C. (IE), Xalapa, Mexico, and the Museum national d'Histoire naturelle (MNHN) Paris, France.

Sixteen third-instar larvae of *L. mazama* were collected on July 13, 1996, 2 km to the south of Yepachic (Chihuahua), in a large oak *(Quercus)* stump, in a mixed pine-oak forest, at an altitude of 1,660 m. The larvae were maintained alive in a terrarium at a relative humidity of 80 percent and a temperature of 23.3°C ± 3.1, supplied with abundant wood and decaying detritus of oak and *Bursera simaruba,* until pupation occurred and the imagos emerged.

A male that emerged November 29, 1996, was dissected June 16, 1997, and a female that emerged November 3, 1996, was dissected November 21, 1996, to examine their reproductive apparatus, which were maintained in Ringer saline solution, fixed in AFATD (ethyl alcohol 96°–formaldehyde–trichloroacetic acid–dimethylsulfoxide), and stained in toto with Feulgen-green light. Some organs were included in Celoidina®-Parafina Histosec®, and histological sections of 7μm were stained using the technique of PAS-Heidenhain hematoxylin (Martínez 1999). Cuticular structures were macerated with potassium hydroxide and stained with chlorazol black (Carayon 1969).

The rest of the adults, two males and three females, were kept as separate male-female pairs, in two terraria with decaying wood, at a relative humidity of 80 percent and a temperature of 25.8°C ±1.8, from February 1997 until their deaths in May 1998. From among these insects, a male, which had emerged November 27, 1996, was selected for dissection after it had copulated, an event that occurred on May 15, 1998.

The anatomical terms used here to describe reproductive apparatus are those proposed by Snodgrass (1933) and Tuxen (1970).

Material Examined. Mexico: coll. E. Borel/ 2 males (MNHN-Paris); /Höge/ex coll. Bolieau/ 1 female (MNHN-Paris); /Höge/ 1 male (MNHN-Paris); Chihuahua: /Santa Clara/Höge/Ex Musaeo H. W. Bates 1892/ 1 male, 1 female (MNHN-Paris); /Santa Clara/Höge/ex coll. Bolieau/ 1 male (MNHN-Paris); Chihuahua: 2 km south of Yepachic, 13-VII-1996, P. Reyes and D. W. Edmonds, coll./ altitude 1,660 m, mixed pine-oak forest, in a decaying oak log/ 2 males, 3 females, 5 third-instar larvae (IE-Mex); 2 km south of Yepachic, 12-VII-1996, P. Reyes and D. W. Edmonds, coll. /altitude 1,660 m, mixed pine-oak forest, in a rotting oak log/ 1male pupa, 1 female pupa, and 3 males and 4 females that emerged in the laboratory during October and November, 1996 (IE-Mex); Sonora: /Yécora/15-VII-1966/P. Reyes and W. D. Edmonds, coll./altitude 1,400 m, by light, between 21:00 and 23:30 hours/ 1 male (IE-Mex); /Puerto de La Cruz, 10 km east of Yécora/17-VII-1996/P. Reyes, coll./altitude 1,770 m, in a rotting oak log/1 adult remains, 1 third-instar larva (IE-Mex).

RESULTS

Distribution and habits. On the dates indicated above, adult males and females and larvae were collected from decaying oak (*Quercus* spp.) stumps and logs that are found in mixed pine-oak forests between the altitudes of 1,400 and 1,770 m in the Sierra Madre Occidental, in the states of Chihuahua and Sonora. One male adult was attracted by light in July. On being collected, adults showed thanatosis and the larvae stridulated.

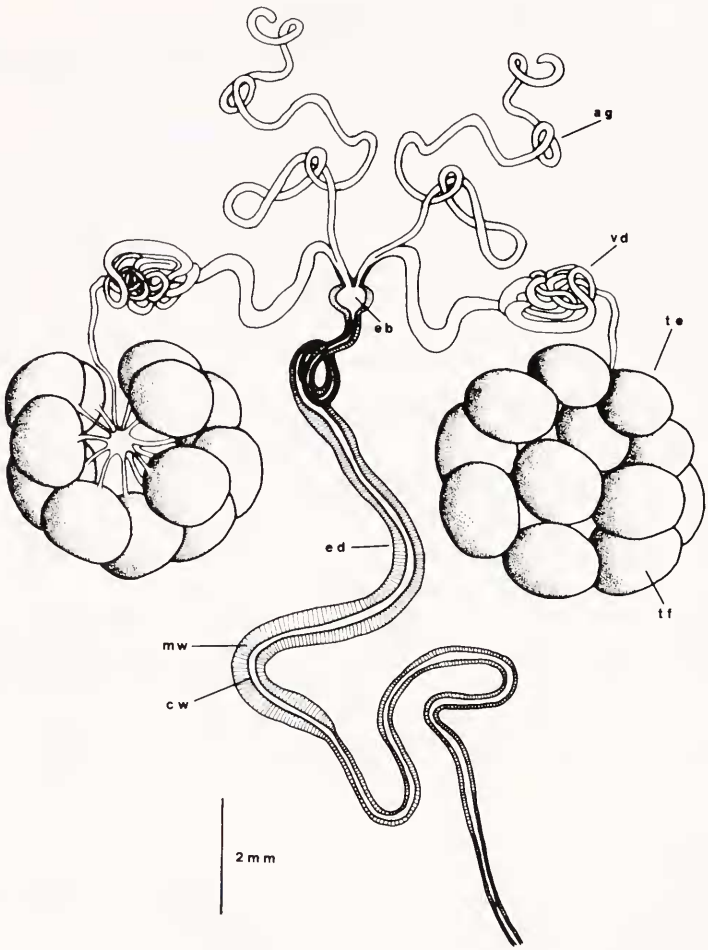

Fig. 1. Scheme of male *Pseudolucanus mazama* reproductive apparatus (ag, accessory gland; cw, cuticular wall of the ejaculatory duct; eb, ejaculatory bulb; ed, ejaculatory duct; mw, external muscular wall of the ejaculatory duct; te, testis; tf, testicular follicle; vd, vas deferens).

Of a total 16 third-instar larvae maintained live in the laboratory (at a relative humidity of 80 percent and temperature of 23.3°C ± 3.11), 7 died, while 9 developed to the pupa stage; of the latter, 7 reached the adult stage (3 males and 4 females). The larvae remained buried the entire time, active in the wood and decaying detritus that served them as both food and substrate. Pupation occurred in the detritus, apparently without construction of a cocoon. In 5 larvae, the prepupa-pupa period lasted from 5 to 8 days. In 7 larvae, the period from pupa to

adult lasted between 31 and 35 days. Adults showed surprising longevity in the laboratory (maintained at a relative humidity of 80 percent and temperature of 25.8°C ± 1.8), up to 18 months, during which time they showed little activity and almost no feeding. In early May 1998, one pair appeared to be preparing to mate and the female fed on dead wood. The adults spent most of their time buried between the terrarium floor and the bottom of the decaying log.

Male reproductive system. The male reproductive system consists of 2 testes, each with 12 testicular follicles, and 2 vas deferentia, 2 accessory glands, an ejaculatory bulb, and an ejaculatory duct (Fig. 1).

The reproductive structures of a young male that had emerged six and a half months prior to study and which was dissected on June 16, 1997, were surrounded by abundant fat-body and tracheoles. In contrast, the male dissected 18 months after emergence, on May 15, 1998, did not show similar fat-body material, but did show abundant tracheoles, and its testicular follicles were smaller than those of the younger male.

Testicular follicles of both younger and older males were free, without a covering membrane, and spherical, though slightly flattened in the dorsoventral direction. In histological observation, the follicles are radial. In the young male, the testes were immature, showing only primary and secondary spermatocyte cysts, and not fully developed spermatozoa. In the 18-month-old male that had copulated shortly before dissection, the histological structure of the testicular follicles showed degenerating cysts without a trace of spermatozoa (Fig. 2). Most likely, spermatogenesis had not occurred, and copulation had not been functional.

A relatively long vas efferens emerges from each testicular follicle. All the vas efferentia flow into the vas deferens of their respective testicle. The vas deferentia show three different regions: a straight region on leaving the follicles, a volu-

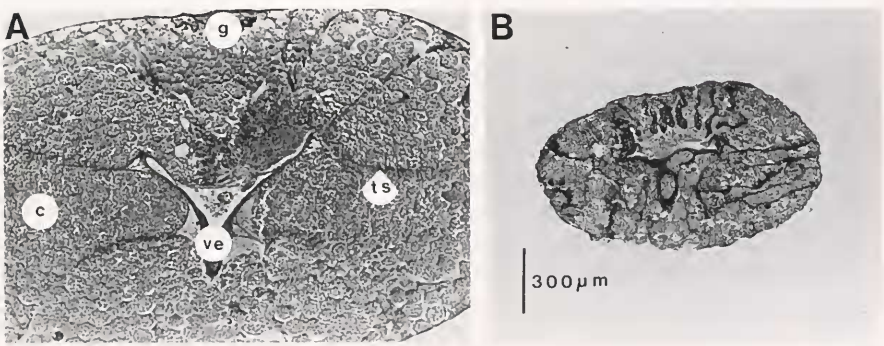

Fig. 2. Microphotographs of histological sections of *Pseudolucanus mazama* testicular ollicles: (A) in a recently emerged male, (B) in a male one year after emergence (c, cysts; g, germarium; ts, testicular septa; ve, vas efferens). The same scale is used in both photographs.

minous region in which they curl around themselves, and an enlarged region that narrows near emergence in the ejaculatory bulb.

The accessory glands are tubiform, relatively long, and rolled around themselves. They both discharge, parallel to the vas deferentia, in the anterior part of the ejaculatory bulb.

The ejaculatory bulb is very small and thick, almost spherical. It is formed of a relatively thick cuticular intima, surrounded by a muscular cloak that is not very thick. The 2 vas deferentia and 2 accessory glands open into the anterior region of the ejaculatory bulb. The ejaculatory duct emerges from the ejaculatory bulb's posterior region.

The ejaculatory duct is very long and has two distinct regions of about the same length. The anterior part, emerging from the ejaculatory bulb, has a cuticular intima covering the duct and a thick muscular wall toward the exterior. The posterior section narrows; while the interior diameter of the duct remains the same as in the anterior region, the duct's muscular wall here is much thinner. This posterior section gives into the aedeagus, which has a permanently everted internal sac with a long terminal flagellum.

Female reproductive system. The female reproductive system is formed of two ovaries, each with 12 ovarioles, and two lateral oviducts, a common oviduct, the bursa copulatrix, spermatheca with its gland, and vagina (Fig. 3).

The entire reproductive apparatus is surrounded with abundant fatty material and numerous tracheoles, to such a degree that during dissection it was difficult to distinguish the different structures. These structures became clearer after being stained in toto with Feulgen-green light, or after maceration and staining of the cuticular structures with chlorazol black.

The female studied had recently emerged and was immature. Each telotrophic ovariole presented a germarium, a vitellarium without oocytes, and a long pedicel (Fig. 4). The pedicels of the ovarioles of each ovary ended in a calix. In each ovary, the ovarioles and calix were enveloped in a membrane. Each ovary's calix continued until it met the corresponding lateral oviduct. Both lateral oviducts terminated in the common oviduct, which was relatively long and surrounded by abundant muscular tissue, and which terminated in the vagina.

The spermatheca was a large, sclerotized structure, formed by the spermathecal capsule, a duct, and a gland. The spermathecal capsule was heavily sclerotized and surrounded by abundant glandular tissue. The cuticular spermathecal duct was surrounded by a thick coat of muscular tissue that emerged from the base of the capsule. The duct's posterior region was thicker and had a folded wall, joining the anterior part of the vagina between the common oviduct and the bursa copulatrix. The spermathecal gland was cuticular and surrounded by glandular tissue that was not very thick; the gland's duct was short, ending in the base of the spermathecal capsule.

The bursa copulatrix had a highly folded cuticular intima, surrounded by a muscular wall. Before terminating in the anterior part of the vagina, the posteri-

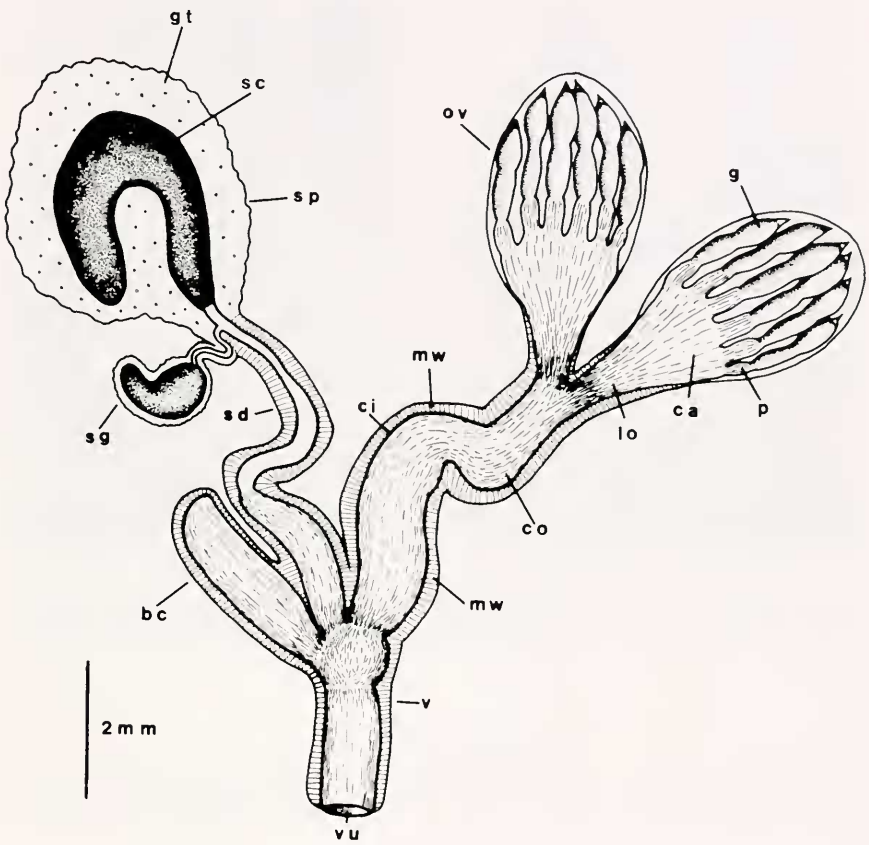

Fig. 3. Scheme of female *Pseudolucanus mazama* reproductive apparatus (bc, bursa copulatrix; ca, calix; ci, cuticular intima seen through the muscular wall; co, common oviduct; g, germarium; gt, glandular tissue covering the spermathecal capsule; lo, lateral oviduct; mw, muscular wall; ov, ovary; p, pedicel; sc, spermathecal capsule; sg, spermathecal gland; sp, spermatheca; sd, spermathecal duct; v, vagina; vu, vulva).

or section of the bursa copulatrix leaned against the base of the spermathecal duct. At this point, these two structures were enveloped by the same coat of muscular tissue.

The vagina's cuticular intima was highly folded and surrounded by a thick coat of muscular tissue. The vagina comprised two different regions. The anterior section was bulky. The common oviduct joined this section toward its beginning, while the bursa copulatrix and the spermatheca joined this section lateral-

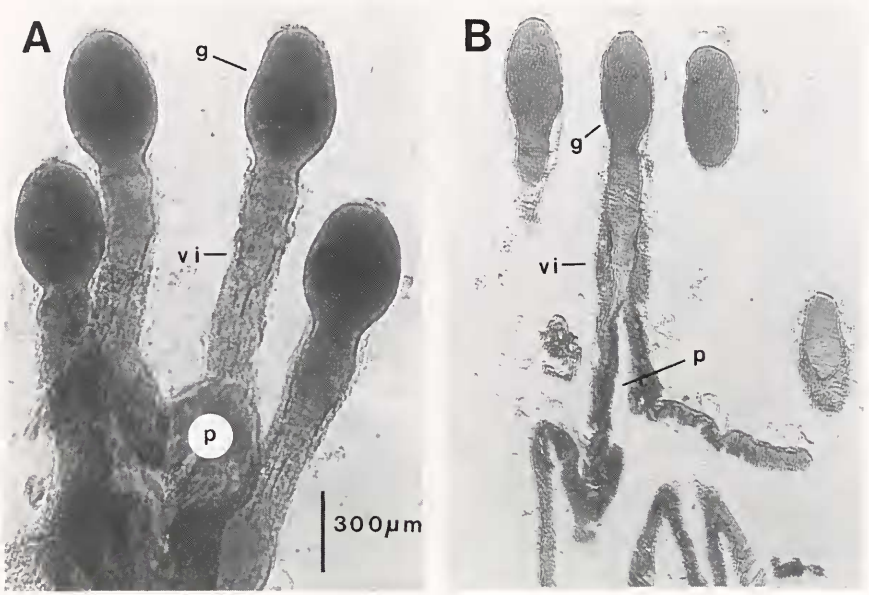

Fig. 4. Microphotographs of *Pseudolucanus mazama* ovarioles: (A) in toto, (B) in a histological section (g, germarium; vi, vitellarium; p, pedicel). The same scale is used in both photographs.

ly. The posterior part of the vagina was straight, terminating in a vulva that opened to the exterior.

DISCUSSION

Lucanus mazama belongs to a genus with wide holarctic and oriental distribution. The genus's 63 species include 5 known in North America; of these, *L. mazama* alone reaches the north of Mexico (Maes 1992). *L. mazama* distribution in Mexico occurs in woods of boreal origin, a mix of pine and oak, in the Sierra Madre Occidental. The species maintains holarctic affinities, being adapted to colder weather, and in the Mexican zone of transition shows a nearctic dispersal pattern (*sensu* Halffter 1976).

In Mexico, *L. mazama* lives in rotting stumps and logs, with a preference for oak (*Quercus* spp.). Both adults and larvae are commonly found in these habitats. The adults are sometimes attracted to light. These characteristics are shared by two related North American species, *L. capreolus* and *L. placidus* (Say 1825) (Ritcher 1966, Mathieu 1969), though the larvae of these two latter species appear more polyphagous. The surprising longevity found in nonfeeding *L. mazama* adults—18 months in the laboratory—contrasts with the longevity of *L.*

capreolus, which has been observed to live in the laboratory for just 3 months with a diet of sugared water provided through a piece of cotton (Mathieu 1969).

The present study provides the first complete description of *L. mazama* reproductive apparatus, both male and female.

In the male, the reproductive anatomy resembles that of *L. capreolus,* studied by Williams (1945), with equal numbers of testicular follicles (12-12), and also that of *L. cervus,* which has 10-10 testicular follicles, and of *Dorcus parallelipipedus,* which has from 10-10 to 12-12 testicular follicles, as studied by Bordas (1900). These two authors described seminal vesicles in these species, but in *L. mazama* we observed no dilation of the vas deferentia that would respond to such seminal vesicles. Lack of seminal vesicles is a common characteristic of other Scarabaeoidea species, notably those of the dung beetle subfamilies (Pluot-Sigwalt and Martínez 1998). The aedeagus of *L. mazama* agrees with that of other Lucaninae (Holloway 1960) in having a permanently everted internal sac and basal piece that does not surround the median lobe. The permanently everted internal sac in *L. mazama* terminates in a long flagellum.

We have no detailed description of female reproductive anatomy in Lucanidae species, except the general scheme of Dorcinae put forward by Franciscolo (1997). This scheme is generally consistent with our observations on *L. mazama,* which shows 12-12 ovarioles, the same number found by Ritcher and Baker (1974) in a female of this species from Arizona, in *Sinodendron rugosum* Mannerheim 1843, and in other Scarabaeoidea (Rutelinae and Cetoniinae).

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